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RESEARCH ARTICLE

A study of the effects of green stem syndrome on some quality parameters in soybean and the possibility of early detection

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ABSTRACT

Green stem syndrome is one of the major problems encountered in soybean production in the world because it makes harvesting with a combine harvester difficult. Although the prevalence of the green stem syndrome Turkey is unknown, in recent years it has started to be observed frequently. Leaf color characters in the growing stages of some soybean varieties have been determined according to varieties in this study. Color changes in the leaves from V3 to R8 phase were monitored using L*, a*, b* color scale. Possibility of detecting changes in leaf color before the R8 stage was studied. Some quality parameters have been evaluated in seed samples obtained from plants with and without symptoms in the R8 stage. It was determined that the germination rate of the seeds obtained from the plants with the syndrome decreased by 61.4% on average compared to those from healthy plants. Furthermore, compared to non-symptomatic seeds, symptomatic seeds were larger, had a lower fat ratio, lower palmitic and linoleic fatty acid values, and higher oleic fatty acid values. At this study was determined that the most significant difference was manifested in terms of stem moisture values during germination and harvesting. In addition, detection of green stem syndrome can be used b* color value as a marker. The hypothesis of the study is that the syndrome can be diagnosed at early stage by following color values in the soybean leaves. In the future studies the color of the leaf can also be a parameter available for the machine learning models.

Keywords: Harvest stage; *Glycine max* (L.); Green stem syndrome; Leaf color; Machine learning

INTRODUCTION

Soybean [Glycine max (L.)] is one of the most widely planted oil seed plants covering an area of 58% in the world. It is an important source of fat and protein as human food and animal feed (Yin et al. 2011). Soybean also contains sugar and phenolic compounds as well as fat and protein (Bellaloui, 2012). The soybean enriches the soil in terms of nitrogen and saves on fertilizer and increases the yield of the following crop making it a good rotation plant (Anonymous, 2018a).

In the cultivation of soybean, Green Stem Syndrome (GSS), also known as the Greening Syndrome, can be encountered in a whole field, a section, in parts of a whole area or scattered individually. The syndrome was initially detected in the 1950s in the USA and thought to be caused by insecticides, however subsequent studies indicated that this was not the case (Daugherty et al. 1964). In those days defoliating agents were used in syndrome infested soybean areas to dry leaves that had remained green as a solution to ensure that combine harvesters could harvest without problems and prevent increasing losses. Subsequently, it was determined that these chemicals had a negative effect on germination and seed quality. And the practice was prohibited (Dale and Walters 1985). In a study carried out by Rabedeaux et al. (2005) in Virginia, it was reported that the syndrome increased harvest losses and seed yields incurred a loss of at least 25%. Egli and Bruening (2006) stated that the syndrome increases as the number of pods decreases; however, they stated that this situation is not stable. Furthermore, it has been emphasized that in addition to decrease in yield, the syndrome affecting the seed quality can be related to insect damage, soil tillage, fungicides, herbicides and water lacking. However, the same results could not be obtained every year (Leonard et al. 2011). In a study carried out by Isobe et al. (2014) in Japan, it was reported that the syndrome is related to genotype and

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location. Fujii et al. (2015) argued that the location is not directly effective with the incidence of syndrome, that the most effective factor may be early flowering. In a study carried out in the USA, 77% of soybean farmers reported that they observed the syndrome during the harvest period on 43% of cultivated areas (Harbach et al. 2016a). Between 2009 and 2012, 86 trials were established in 7 different locations in the US and it was concluded that the syndrome emerged as a result of genetic and environmental interactions (Harbach et al. 2016b). Hajong et al. (2016) emphasized that the causal agents of the syndrome were not fully known and therefore fighting it was a challenge. In the study, it was determined that with warm and dry weather conditions exceeding normal seasonal conditions the disease increased. They also determined that the green stems, green leaves and high seed moisture during harvest increased harvest losses in the combine harvester.

Although the ratio for soybean is not known under main product conditions in Turkey where soybean can be grown as a main crop as well as a second crop, it has been noted that the number of green leaves and stems of soybean plants increases during the R8 phase when the pods reach maturity. This syndrome is not yet known by the producers and therefore this condition is defined as a genotype property by them.

There is scarcely any study that has been carried out in Turkey which focus on the impact of GSS on the quality and yield of soybeans. The studies conducted on GSS are generally as a 0-5 scale evaluation used in plant breeding studies. Arslan and Arıoglu (2003) used 13 varieties in the maturity group V suitable for Amik Plain in a study involving soybean variety adaptation as a second crop and reported that GSS was observed in US origin varieties. However, it was emphasized that the factors could not be determined. In a study carried out by Cubukcu (2017), it was reported that the rate of GSS in soybean cultivation areas in Cukurova Region has increased. At study was evaluated the effect of GSS on varieties and lines according to the 0-5 scale by Cubukcu (2017). As a result of the evaluation, it was determined that the syndrome had incurred CU04-74, CU03-75-1 the most level while had incurred CU04-122, Sa-88, CU04-45, Adasoy, CU05-13 medium level.

Color is one of the important distinguishing features used to determine the quality and quantity of plants. Generally, it is used in the determination of the chlorophyll content of plant leaves, mineral matter deficiencies, water demand, plant quality or stage. In addition, it is used in determining the effect of some biotic and abiotic stress on the plant growth in the monitoring of diseases or disease levels in plants, as well as in monitoring and predicting diseases (İnce and Vurarak, 2019). Rantnasari et al. (2014) reported that disease symptoms manifesting in 15% of sugar cane leaves affected the yield and developed a model that could be used to predict the disease with color change. In addition, researchers determined the digital image of the diseased leaves and L*, a*, b* color model and reported that plants could be determined with an accuracy rate of 80% before they became infected.

Contributing to the use of color following in the field of machine learning is one of the aim of the study. If there is relationship between color and symptom, this data can also be used to machine learning in subsequent sutudies. Leaf color characters in the growing stage of varieties of healthly and GSS soybean plant have been determined in this study. Furthermore, the possibilities of detecting the syndrome before the R8 growing stage (harvesting stage) have been investigated. Color changes in the leaves of soybean were followed according to different lines/varieties and different growing stages. Furthermore, samples taken from all varieties of healthy and GSS soybean plants were investigated in terms of leaf color, germination, 1000 seed weight, protein content, fat content and fatty acids in laboratory conditions.

MATERIALS AND METHODS

Materials

The trials were carried out in the trial areas of the Eastern Mediterranean Agricultural Research Institute (EMARI) in Adana/Turkey. The soil of the experiment field was at an altitude of 12 m consisted of 29.1-50.4% silt, 11.5-55.3% sand and 19.8-39.2% clay. Lime was 12-20%, organic matter was between 1.22-2.58%. CEC (cation exchange capacity) was between 21.32-34.76 cmolkg^{-1}, pH 7.49-7.92. Available phosphorus (P_2O_5) values in soil were determined between 3.1 and 17.8 kg^{-1}. According to climate data for Adana province, the highest and lowest temperatures were recorded as 29°C in August and 21°C in May (long term climate data), respectively. The most humidity in the province occurs in May. Fig. 1 displays the climate data recorded during the 2017 experiment period.

Some technical characteristics of soybean lines/varieties used as materials in the study are given in Table 1. Out of these lines and varieties Nazlıcan, Türksoy, Adasoy and Yemsoy have been developed by EMARI in the climate conditions of Adana.

This study was conducted between May-October 2017 under main crop conditions. The sowing was done by using 8 kg da^{-1}. The seeds were sown in fourth week of April via pneumatic precision drill and with 70x5 cm distance.

Table 1: Some climatic data recorded during the experiment (2017)

Months	Temperature (°C)			Other climate data		
	Max.	Ave.	Min.	Humidity (%)	Wind (km/h)	Precipitation (mm)
April	35	23	12	63	11	38.87
May	33	21	11	70	10	109.7
June	37	25	14	68	9	28.96
July	43	29	19	63	10	0
August	43	29	21	66	9	0
September	38	26	16	64	9	1.05
October	32	22	12	51	10	28.95

*http://mobile.wunderground.com/history/airport/LTAG/2017/10/31/DailyHistory.htm

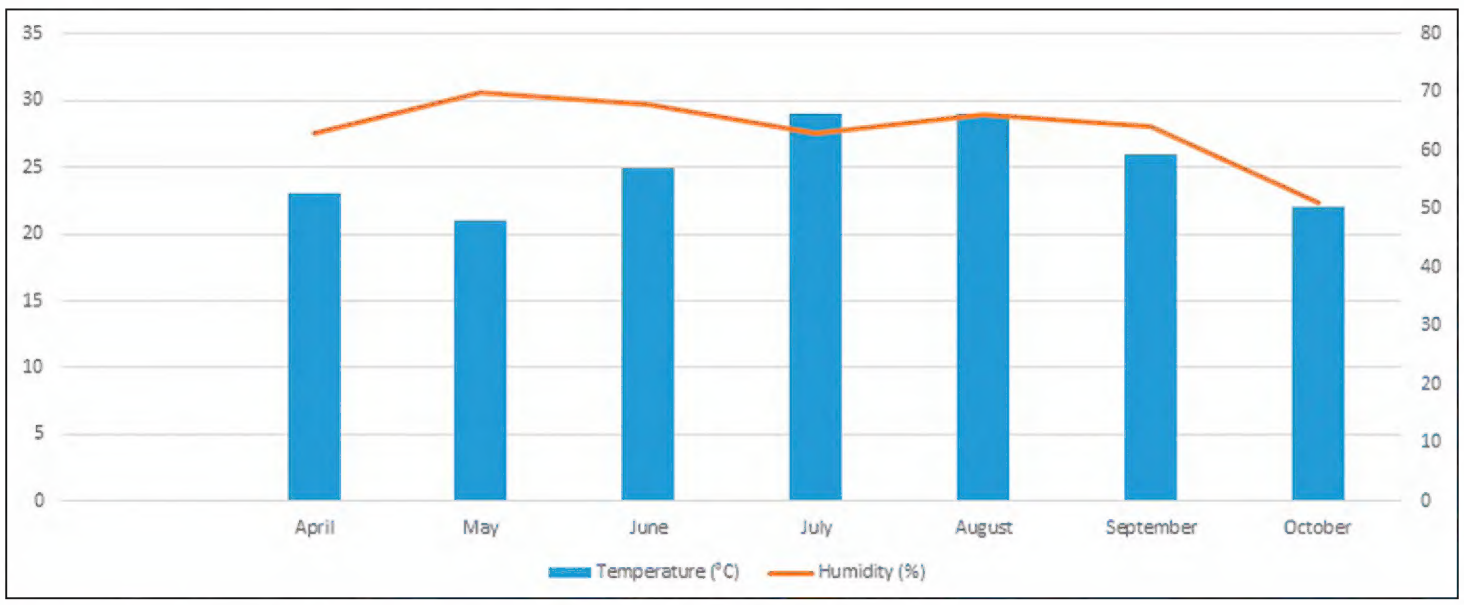

Fig 1. Temperature and humidity values at the growing time of soybean in 2017

The seeds were inoculated with bacteria before sowing. And than 3 kg da-1 pure nitrogen and 6 kg da-1 pure phosphorus were used as fertilizer during growing season. The plants were watered in total 4 times depending on the water demand of the plant with an interval of 15-20 days. While the leaves and stems of soybeans with GSS remain green at harvest time, the pods of soybeans without GSS are fully mature at harvest time (Fig. 2a-2b).

Methods

Field Experiments were designed as a randomized block with three replications. The parcels were designed to be 5 m x 2.8 m. In the seed samples taken from the healthy and GSS infested plants determined in each plot in the R8 (harvest stage) in the experiment to analyses for 1000 seed weight, protein-oil ratios, composition of fatty acids and stem moisture. This laboratory analyses were designed as a split plot design (varieties as main plots and growing stages as subplots) with three replications. The main plots were designed 11 lines/varieties (Ceysoy, Blaze, Ataem-7, CU04-122, Atakisi, Sa-88, Cinsoy, Nazlican, Türksoy, Adasoy, Yemsoy). And the sub-plot consisted of 5 stages of grow (V3, R2, R4, R6, R8). The 5 stages in which leaf samples were taken include vegetative and generative periods. The V3 stage of these phases includes four internodes and three genuine leafy periods and the plants are 18-23 cm tall. Stage R2 is defined as the stage of full flowering and is the stage where there is a flower and a fully formed leaf in any of the top two internodes on the main stem and plant height is 45-55 cm. The R4 phase is a rapid pod formation stage. One of the last four internodes on the main stem has a 2 cm long pod. The phase R6 is the phase of maximum seed formation. One of the last four internodes on the main stem is a pod with a green seed that fills the inside of the fruit. R8 stage is the harvest maturity phase and 95% of the pods taken on the color of maturity (Fehr and Caviness 1977; Williams et al. 1999).

Determination of color values (L, a*, b*)*

In the five growing stages determined, the leaf samples were collected in a random order (from the lower, the middle and the upper parts) in such a way as to have three replications of each kind which were mixed and shredded through a blender. Minolta CR-5 Chromameter (Osaka, Japan) colorimeter was used for color determination of the leaves samples. The colorimeter device was standardized against a white ceramic plate before each use. L*, a*, b* values are given by a 3D coordinate system and L* value in this coordinate system determines the direction from brightness to darkness in a vertical axis. The L* value ranges from 0 to +100, with 0 representing blackness, +100 whiteness. a* indicates the change to + a* red, -a* to green. This value is between -90 and +90. The b* value indicates the change from + b* to yellow and -b* to the blue. This value is between -90 and +90 (McGuire, 1992; Lee and Castle, 2001).

The crude protein content (calculated as N x 5.71) was determined on soybean samples by the standard Kjeldahl procedure. A Turbothem (Gerhardt, Germany) and a Gerhardt distillation unit, Vapodest 45 S, were used for the analysis. The sample size used in the Kjeldahl procedure was around 1.00 g. Samples were weighed and transferred in to Kjeldahl digestion flask containing catalyst (mixing of K_2SO_4 and $CuSO_4H_2O$) and 12 mL of concentrated H_2SO_4. After 2.5 h of digestion in a unit with electrical heat and füme removal and cooling to room temperature 40 mL of NaOH base (mass fraction w = 33%) was added to each flask. By distillation, ammonium hydroxise was trapped as ammonium borate in a boric acid solution. The initial pH of distillation was measured by Titroline. Total nitrogen determined by titration up to initial pH with standardized HCL (AOAC 2005). Crude oil was extracted with petroleum ether using a Soxhlet apparatus for 4 hours. Determination of oil content of soybean samples soxhelet automatic oil in the extraction device (Gerhardt, Soxtherm 2000) carried out using petrollium ether (AOAC 2005). The samples (5 g) were extraced with 140 mL of petrollium ether at an extaction temperature of 150°C for 80 minutes. Fatty acid methylesters obtained by methylation of total lipids were analysed by the American Oil Chemists' Society according to the method described in the analysis methods of Ce1-62 (AOCS 2005).

1000 seed weight (g)

It was obtained by multiplying the average of the weights of 100 seeds taken in 4 parallel with 3 replicates from the parcels of each type with 10 (Anonymous, 2010).

Germination rate (%):

9 cm diameter petri dishes were used for the germination test, and 25 seeds were planted in each petri dish. Germination tests were carried out in a dark environment in germination cabinets set at 25 ± 2°C. (Arif et al. 2002).

GSS scale assessment

Evaluation scores of 0-5 were given based on the observations made in terms of the varieties. The percentage of these points determines the percentage of the disease in terms of the varieties. According to the scale values, 0 equals no disease, 1 indicates a disease level of 1-10%, 2 is commensurate with a rate of 11-35%, 3 means 36-65% while 4 corresponds to 66-90% and 5 is 91-100% (Little and Hills 1978).

The data obtained in the study were subjected to variance analysis using a statistical package program. Parameters

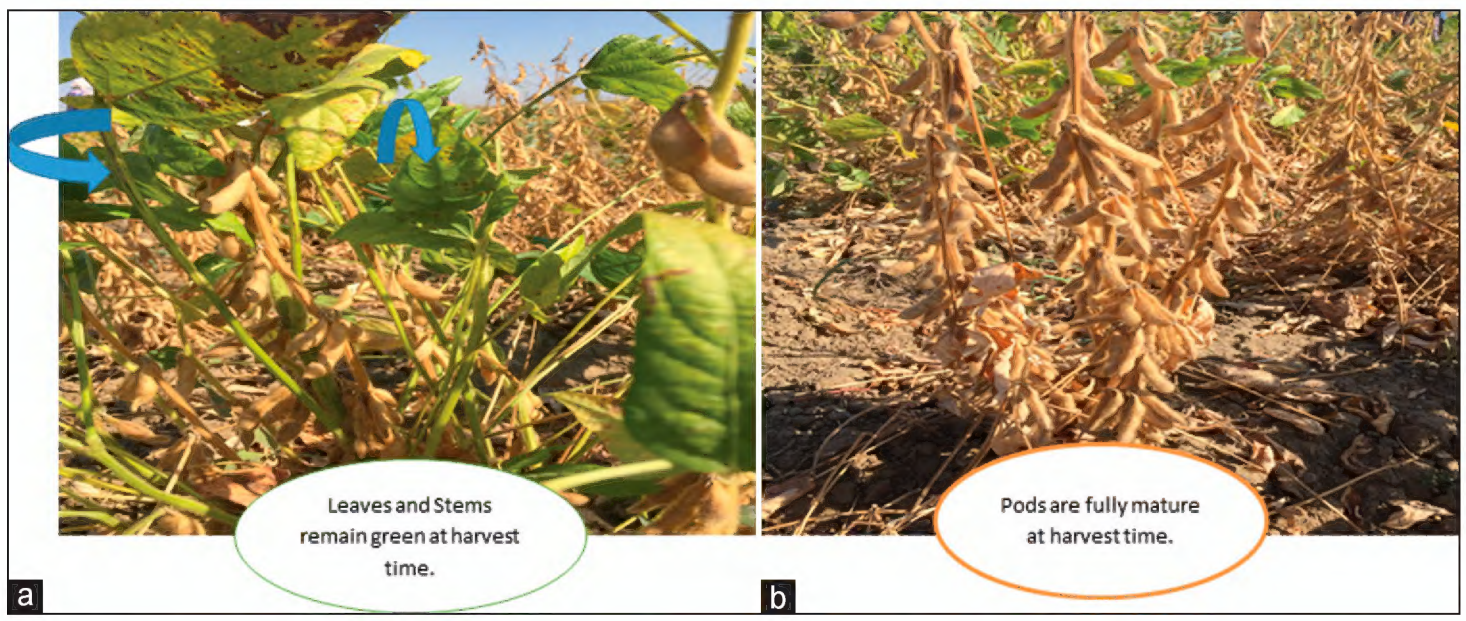

Fig 2. (a) GSS observed soybeans; (b) Non GSS observed soybeans

Table 2: Catalog data of some technical characteristics of lines and varieties*

Line/varieties	Potantiel seed yield (kg da^{-1})	Plant height (cm)	Leaf form	Flower color	Protein rate (%)	Fat rate (%)	1000 seed weight (g)	Breeding region
Nazlıcan	400-450	120-150	spear	violet	33-35	20-22	180-210	Turkey-Adana
Türksoy	380-440	125-160	ovate	violet	31-24	20-23	160-180	Turkey-Adana
Adasoy	400-450	120-150	oval	white	33-36	22-24	140-170	Turkey-Adana
Yemsoy	350-400	120-150	oval	violet	32-34	18-20	180-210	Turkey-Adana
Ceysoy	330-430	139-153	oval	violet	39-41	22-24	116-200	Turkey-Adana
Blaze	350-375	98-130	oval	violet	39-40	20-23	138-218	abroad
Ataem-7	300-360	133-157	oval	white	38-42	21-22	138-230	Turkey -Antalya
CU04-122	350-400	127-153	ovare	violet	41-43	20-22	116-183	Turkey-Adana
Atakişi	350-380	119-137	oval	white	38-40	22-23	115-187	Turkey-Adana
Sa-88	300-350	120-137	oval	violet	37-39	22-23	100-160	abroad
Cinsoy	350-450	115-130	oval	violet	39-40	21-23	127-221	Turkey -İzmir

*Source: (Anonymous, 2017; Anonymous, 2018)

that indicated significant F values at a significance level of 5% were subjected to multiple comparisons with 'LSMeans student's t' test (Düzgünes et. al. 1983).

RESULTS AND DISCUSSION

Table 2 shows the results of GSS evaluation according to the 0-5 scale from the counts made during phase R8. According to these scale, Green Stem Syndrome was observed in Ataem-7 variety in the range of 91-100%, Blaze, Atakişi, Sa-88 varieties depicted syndrome symptoms at a rate of 66-90%. The varieties displaying the least syndrome symptoms with a scale value of 1 were the Nazlıcan, Türksoy, Adasoy and Yemsoy varieties. These four varieties have been developed within the scope of breeding programs made in the test fields of EMARI. In this respect, it is possible to say that this syndrome can be seen more in varieties developed under different environmental conditions.İt is thought that the locations where the cultivars were brend may be importent in terms of the frequency of symptoms.

In studies conducted in Japan, it has been reported that the syndrome is related to the genotypes and locations where soybean is grown (Isobe et al. 2014; Fujii et al. 2015). However, they argued that the most effective factor was early flowering. According to Table 2, the Sa-88 variety has the earliest flowering day with 28 days and with the GSS rate of 66-90%. On the other hand, Türksoy variety which ranks with 29 days in terms of flowering has the GSS value of between 1-10%. This result is considered to supports the conclusion that the breeding region of the varieties is more influential than the number of flowering days. Disease symptoms were less noted in early flowering cultivars. However, Blaze, Ataem-7, Atakisi, and Sa-88 with a GSS scale of 4 and 5 have a flowering day average with 39.5 days while Nazlıcan, Türksoy, Adasoy, Yemsoy varieties which have a 1 GSS scala value have a flowering day average with 31.0 days.

Table 3: GSS scale values of lines and varieties in the experiment area and 50% flowering

Line/varieties	GSS scale values*	Observed ratio (%)	%50 flowering (day)
Ceysoy	2	11-35	30
Blaze	4	66-90	33
Ataem-7	5	91-100	31
CU04-122	2	11-35	32
Atakişi	4	66-90	32
Sa-88	4	66-90	28
Cinsoy	2	11-35	33
Nazlıcan	1	1-10	34
Türksoy	1	1-10	29
Adasoy	1	1-10	44
Yemsoy	1	1-10	51

*0: %0, 1: %1-10, 2: %11-35, 3: %36-65, 4: %66-90, 5: %91-100 (Little and Hills, 1978).

Table 3 shows that the growing stage and varieties were found to be statistically significant impact ($p<0.01$) on the L* value. The L* value is the highest in the R8 phase harvesting period. It has been determined that the closest phase to this period is the V3 phase. According to the GSS scala, varieties that are included within the 4-5 scala (1-5 disease rating scala) value are Ataem-7, Sa-88, Atakişi and Blaze varieties respectively. The average brightness (L*) of these varieties is 30.94 on average. The brightness average calculated for the other varieties and lines is 29.92. Türksoy and Sa-88 varieties were statistically included in the same group in terms of L* brightness values, however it has been determined that in terms of syndrome frequency, Türksoy has a frequency rate of 1-10% and Sa-88 a frequency rate of 91-100%. According to these results, it can be said that the probability of detecting the syndrome is by following the L* brightness value in the early stages is low. a* value which is used for color tracking from red to green showed significant differences ($p<0.01$) in the comparison of stages and it was determined that the value of a* value changed from green to red as the plant progressed from phase V3 to phase R8. However, it has been determined that the changes in the a*

Table 4: Variance analysis table of color value changes according to phase and line/varieties

Parameters	L*	a*	b*
Growth phase (E)			
V3	31.57^{b}	-5.26^{b}	14.47^{b}
R2	29.85^{c}	-4.80^{b}	11.36^{e}
R4	24.88^{d}	-5.52^{b}	12.88^{d}
R6	29.91^{c}	-5.35^{b}	13.68^{c}
R8	35.28^{a}	-3.37^{a}	20.36^{a}
$LSD_{.05}$	0.64	0.84	0.53
line/varieties (HC)			
Ceysoy	28.47^{e}	-4.33	15.83^{ab}
Blaze	29.95^{cd}	-4.37	14.37^{e}
Ataem-7	31.25^{ab}	-5.21	16.03^{a}
CU04-122	30.75^{bc}	-4.44	15.08^{b-e}
Atakişi	30.71^{bc}	-5.36	15.21^{b-d}
Sa-88	31.86^{a}	-5.34	14.84^{c-e}
Cinsoy	30.86^{bc}	-5.34	14.61^{de}
Nazlıcan	29.28^{de}	-4.40	15.42^{a-c}
Türksoy	31.92^{a}	-5.46	15.34^{a-d}
Adasoy	28.84^{e}	-4.25	14.72^{c-e}
Yemsoy	29.32^{de}	-4.98	15.22^{b-d}
$LSD_{.05}$	0.96	-	0.79
CV (%)	4.38	35.40	7.24
p value			
E	<.0001**	<.0001**	<.0001**
HC	<.0001**	0.2621^{ns}	0.0014**
E*HC	<.0001**	0.2445^{ns}	<.0001**

*, **, ns significant at the levels of 5%, 1%, and not significant respectively; Means in each column with the same letters are not significantly different ($p<0.05$)

value according to varieties is not statistically significant. It has been concluded that a* cannot be used as a marker for early diagnosis of the syndrome. It was determined that the b* value, which is used to monitor color change from yellow to blue, manifested differences on a statistically significant level ($p<0.01$) according to the stages. It was determined that the highest level of brightness was observed in the R8 phase and this phase was followed by the V3 stage. According to the GSS scale, the highest level of green stem syndrome was found in the Ataem-7 variety which also displayed the highest b* values compared to the other varieties. It can be concluded that in terms of using color change to monitor the syndrome, the b* is a more appropriate color parameter to be used as a marker than the L* and a* values.

Table 5: The results of the variance analysis of changes in some quality parameters of seeds of healthy and GSS infested plants

Parameters		GSS plants samples			Healthy plants samples		
		p value	$LSD_{.05}$	CV (%)	p value	$LSD_{.05}$	CV (%)
1000 seed weight	(g)	<.0001**	0.84	2.5	<.0001**	1.67	5.2
Fat ratio	(%)	<.0001**	0.85	2.2	0.0244*	1.02	2.6
Protein ratio	(%)	<.0001**	1.41	2.1	<.0001**	1.89	2.8
Palmitic acid	(%)	<.0001**	0.52	2.9	<.0001**	0.28	1.5
Stearic acid	(%)	<.0001**	0.32	4.6	<.0001**	0.21	3.2
Oleic acid	(%)	<.0001**	0.64	1.4	<.0001**	0.63	1.5
Linoleic acid	(%)	<.0001**	1.29	1.5	<.0001**	0.85	1.0
Germination	(%)	<.0001**	16.0	20.2	0.0018**	21.48	16.3
Stem moisture	(%)	0.0001**	3.41	3.1	0.4121^{ns}	-	13.6

*, **, ns significant at the levels of 5%, 1%, and not significant respectively

Table 6: Change variance analysis of seed quality and stem moisture values taken from healthy and GSS plants according to lines and varieties

line/varieties	1000 seed weight (g)		Protein ratio (%)		Germination (%)		Stem moisture (in harvesting) (%)	
	GSS	Healthy	GSS	Healthy	GSS	Healthy	GSS	Healthy
Ceysoy	20.99^{c}	19.47^{bc}	40.56^{ab}	41.19^{ab}	17.33^{d}	50.00^{e}	65.03^{ab}	20.45^{ab}
Blaze	21.37^{b}	19.40^{bc}	39.67^{b-d}	40.61^{bc}	0.00^{e}	54.66^{de}	63.57^{b}	22.40^{ab}
Ataem-7	22.99^{a}	21.64^{a}	40.76^{ab}	41.18^{ab}	44.00^{c}	82.00^{a-c}	62.17^{bc}	23.33^{a}
CU04-122	18.15^{de}	16.80^{de}	40.09^{a-c}	42.87^{a}	88.00^{a}	93.33^{ab}	64.09^{b}	22.69^{ab}
Atakişi	18.78^{d}	17.90^{cd}	39.54^{b-d}	41.13^{a-c}	46.00^{bc}	68.00^{cd}	68.37^{a}	22.22^{ab}
Sa-88	17.56^{e}	16.11^{e}	38.68^{cd}	37.98^{de}	0.00^{e}	72.66^{b-d}	63.59^{b}	23.14^{a}
Cinsoy	22.71^{a}	19.15^{bc}	41.35^{a}	39.26^{cd}	$4.66d^{e}$	67.33^{cd}	62.64^{b}	20.74^{ab}
Nazlıcan	22.31^{a}	20.40^{ab}	39.57^{b-d}	38.33^{de}	80.00^{a}	88.66^{a-c}	56.61^{d}	22.40^{ab}
Türksoy	18.13^{de}	16.47^{de}	38.51^{d}	37.52^{de}	84.00^{a}	86.00^{a-c}	62.77^{b}	23.33^{a}
Adasoy	17.86^{e}	15.91^{e}	36.60^{e}	37.11^{ef}	90.66^{a}	97.33^{a}	59.02^{cd}	17.85^{b}
Yemsoy	20.17^{c}	17.43^{de}	36.56^{e}	35.41^{f}	61.33^{b}	86.66^{a-c}	63.27^{b}	19.48^{ab}
Mean	20.09	18.24	39.34	39.38	46.90	76.96	62.83	21.64

Means in each column with the same letters are not significantly different ($p<0.05$)

Table 7: Change variance analysis of fat and fat acid ratios from the seeds of healthy and GSS plants according to lines and varieties

line/varieties	Fat ratio (%)		Palmitic acid (%)		Stearic acid (%)		Oleic acid (%)		Linoleic acid (%)	
	GSS	Healthy	GSS	Healthy	GSS	Healthy	GSS	Healthy	GSS	Healthy
Ceysoy	22.90^{b}	23.98^{ab}	10.42^{b}	10.71^{de}	4.23^{bc}	4.09^{bc}	25.55^{e}	24.37^{d}	49.83^{c}	52.88^{c}
Blaze	22.99^{b}	23.25^{bc}	10.42^{b}	11.59^{b}	3.73^{e}	3.64^{e-g}	29.62^{c}	29.13^{b}	46.22^{d}	46.55^{e}
Ataem-7	22.47^{bc}	23.09^{bc}	10.52^{b}	10.96^{cd}	4.20^{cd}	3.89^{cd}	24.54^{f}	21.48^{f}	51.21^{b}	54.19^{b}
CU04-122	22.36^{bc}	22.67^{c}	10.56^{b}	10.97^{cd}	3.82^{e}	3.76^{de}	21.06h	20.78^{g}h	54.30^{a}	54.78^{ab}
Atakişi	23.17^{b}	23.43^{bc}	10.50^{b}	10.83^{c-e}	3.81^{e}	3.73^{d-f}	22.94^{g}	20.99^{fg}	53.10^{a}	55.08^{a}
Sa-88	22.96^{b}	23.28^{bc}	10.41^{b}	11.43^{b}	3.86^{e}	3.53^{fg}	20.88h	20.27h	53.99^{a}	54.96^{ab}
Cinsoy	23.22^{b}	23.51^{bc}	11.16^{a}	11.62^{b}	3.90^{de}	3.66^{ef}	23.58^{g}	23.60^{e}	51.51^{b}	52.40^{c}
Nazlıcan	21.90^{cd}	22.69^{c}	11.11^{a}	12.08^{a}	4.27^{bc}	4.14^{ab}	28.81^{d}	26.85^{c}	46.32^{d}	48.58^{d}
Türksoy	21.40^{d}	23.14^{bc}	9.57^{c}	10.56^{e}	4.71^{a}	4.35^{a}	38.76^{a}	31.28^{a}	40.30^{e}	45.69^{f}
Adasoy	24.30^{a}	24.66^{a}	10.33^{b}	11.11^{c}	3.83^{e}	3.44^{g}	25.90^{e}	23.13^{e}	49.43^{c}	53.04^{c}
Yemsoy	21.84^{cd}	23.86^{ab}	9.29^{c}	10.02^{f}	4.53^{ab}	4.10^{bc}	32.09^{b}	31.45^{a}	45.11^{d}	47.13^{e}
Mean	22.68	23.41	10.39	11.08	4.08	3.85	26.70	24.85	48.96	51.14

Means in each column with the same letters are not significantly different ($p<0.05$)

Image Segmentation is the process that is used to distinguish object of interest from background for the identification of plant diseases. The proposed approach uses CIE L* a* b*, or CIELAB, color scale for use (Kulkarni and Ashwin, 2012). There are also some studies which have been carried out to determine the correlation between quality, disease, developmental period, shelf-life or nutrient content and color. Landschoot and Mancino (2000) carried out a study using color parameters to distinguish grasses from each other and reported that the most important distinguishing indicator for grasses was the hue angle (h°). Deak et al. (2007) reported that there was an association between strawberry leaf color and fruit quality and that light green or yellow leaves during the flowering period had a negative impact on fruit taste. Leon et al. (2007) determined a significant association between hue angle and L* value and chlorophyll in their study and reported that the color meter and chlorophyll content could provide information. Itle et al. (2009) found a high correlation between a* value and total carotene and b* value and lutein in the determination of the carotene values of different pumpkins. Keskin et al. (2013) reported that leaf water content and potassium and calcium content could be determined with high accuracy (R^2> 0.88) with color measurement. Chaudhary et al. (2012) that an algorithm for disease spot segmentation using image processing techniques in plant leaf is implemented. This is the first and important phase for automatic detection and classification of plant diseases. As a results, all these color models are compared and finally 'A' component of CIELAB color model is used.

In the R8 phase before harvest, healthy and GSS plant samples were determined in the plots belonging to the varieties and 1000 seed weight, germination percentage, stem moisture, protein ratio, fat content and fatty acid composition changes were determined according to health and syndrome infestation (GSS plants) status (Table 4, 5 and 6). All parameters except fat ($p<0.05$) and stem moisture (ns) were statistically significant ($p<0.01$) depending on whether the plant was healthy or had GSS. In general, it was determined that in addition to some quality parameters, healthy and GSS infested plants also manifested differences in terms of germination ratio as well as stem moisture (Table 4).

The data obtained from plants with GSS were compared with 1000 seed weight, fat and protein ratios, palmitic acid, stearic acid, oleic acid, linoleic acid, germination rate and stem moisture obtained from healthy plants and 9.2%, - 3.2%, 0.1, - 6.7%, 5.6%, 6.9%, - 4.5%, - 64.1% and 65.6% differences were determined, respectively. According to these results, it was determined that the seeds of GSS plants were larger, fat ratio was low, the protein ratio was almost unchanged and that while palmitic and linoleic acid values were low the stearic and oleic fat acids values were higher than those of health plants. The biggest difference was determined for germination and stem moisture. Especially the differences in fatty acids are thought to have an effect on germination (Tables 5 and 6). It was determined that the highest amount of stem moisture among the varieties in the plants with GSS was Atakişi, while the lowest rate of germination was determined in the Blaze and Sa-88 varieties. The seed of Infected plant have been found o lose 100% of their germination (Blaze and Sa88 variaties). These varieties have followed by Cinsoy with a germination loss of 74.26%. The germination loss of Nazlıcan, Türksoy, Adasoy and Yemsoy varieties with GSS scale 1 (ratio as 1-10%) was calculated as 9.8%, 2.3%, 6.9% and 29.2%, respectively. Therefore, it is possible to say that Türksoy variety is the least affected by GSS in terms of germination loss. The highest rate of germination of seeds obtained from healthy plants was from the Adasoy variety. Furthermore, the stem moisture of this variety was the lowest during the harvest period compared to the other varieties. Türksoy and Sa-88 varieties had the highest stem moisture.

Leonard et al. (2011) reported that GSS affected seed quality in addition to decreased yield. Harbach et al. (2016b) carried out 86 trials in 7 different locations in the USA between 2009 and 2012 and it was determined that there was a positive correlation between the syndrome and yield, plant height, seed color, seed moisture at harvest, protein and fat content during some years, but a negative correlation between rainfall in May, June and July and the frequency of the syndrome.

CONCLUSION

It is believed that the b* value is more usable than the L* and a* values in the L*, a*, b* color value scheme as an indicator to detect or monitor the syndrome in soybean fields. The results may be a starting point of for researchers who want to study machine learning models to identify the GSS in infected soybean during early growing stage of the plants. The use of the a* color value in the predetermination of the syndrome does not appear statistically viable. However, it is necessary to establish testing in the long term in different regions. The germination rate of seeds obtained from plants with the syndrome decreased by 61.4% on average compared to healthy plants and Blaze and Sa-88 varieties were affected in terms of germination rate by 100%. Furthermore, it has been determined that GSS has a negative effect on fat, protein, palmitic fatty acid and linoleic fatty acid ratios. From the results it can be concluded that the varieties affected least by GSS syndrome are Nazlıcan, Türksoy, Adasoy and Yemsoy varieties and

that the rate of GSS infestation decreases in regions where breeding studies of the varieties are carried out.

Author contributions

Yasemin Vurarak; Conceptualization, Methodology, Formal analysis, Writing - Original Draft.

Pınar Cubukcu; Writing - Review & Editing and Project administration.

Ahmet Korhan Sahar; Formal analysis,Writing - Review & Editing.

Celile Aylin Oluk; Laboratory analysis, Review & Editing.

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